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Our study involves predicting infection and symptomatology in humans from psychological status, health behaviors, and immunologic measures taken prior to a viral challenge (Rhinoviruses 2, or 9, or 14). Subjects in the viral challenge trials were 302 volunteers kept in isolation and monitored closely for viral shedding, and symptomatology over nine-day trials. We also collected cross-sectional data on psychological and behavioral characteristics and immune status from an additional 385 volunteers (total of 687 for analyses of these data). ONR funds were used to (a) examine the usefulness of antibody levels to Herpes Simplex Virus 1 (HSV-1) as an indirect measure of cellular immune function in this context; and (b) to examine the influence of smoking status and rate (as assessed by serum cotinine) on immunity and susceptibility to infection.

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19.   HSV-1 antibody levels (IgA and IgG) as measured by ELISA were not correlated with psychological measures in HSV-1 seropostive persons.   Contrary to prediction, higher levels of HSV-1 IgA were associated with less susceptibility among persons challenged with RV2, but not associated with susceptibility for those challenged with RV9 or RV14.   Smoking status and rate (as assessed by cotinine) were associated with nonspecific (across cell populations) increases in numbers of peripheral white blood cells.   This elevation is still detectable one year post-quitting but is no longer detectable five years post-quitting. Smoking status as verified by cotinine constitutes a four-fold increased risk for clinical colds for women but no increased risk for men.   Although women smokers do not differ in rates of infection, they are six-times more likely than women nonsmokers to manifest clinical symptomatology if they are infected.   The increased risk for women (but not men) smokers is found whether the outcome is physician's clinical judgment, total upper respiratory symptom scores, or total mucus weights.

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Overview of Purpose

This report focuses on measures funded by ONR in the context of a larger study designed to determine the roles of psychosocial states, immunologic status, and health practices in individual susceptibility to upper respiratory infection, and clinical colds. On entrance to the study, 687 healthy volunteers between the ages of 18 and 55 completed psychological and behavioral protocols and had blood drawn for immune assessments. This sample is used in this report to examine differences in pre-viral challenge immunity based on psychological states and on smoking status.

Subsequent to the collection of these data, 302 of the volunteers were challenged with one of three rhinoviruses (Types 2, 9, and 14). This sample is used to evaluate the influence of psychological and immune measures assessed pre-challenge on susceptibility to infection and clinical disease. Volunteers in the challenge sample were kept in isolation and monitored closely for viral shedding, and symptomatology over nine-day trials. Serum samples for determination of viral-specific antibody were collected prior to challenge and approximately 28-days post-challenge. The primary outcomes were infection (viral shedding and/or increase in viral-specific antibody) and clinical colds (infection + clinician diagnosis of cold). The primary purpose of the study was to predict infection and clinical colds from measures of psychological states, health practices and immune status taken prior to viral challenge.

This report focuses on two ONR funded aspects of the larger project: (1) examination of the possible usefulness of HSV-1 antibody levels as an indirect measure of cellular immune function, and (2) examination of the effects of smoking status and rate (as biochemically assessed by serum cotinine) on immunity and susceptibility to infection.

HSV as an Indirect Measure of Cellular Immune Function

This work was stimulated by Glaser's argument (e.g., Glaser & Gotlieb-Stematsky, 1982) that latent herpes viruses are held in check by cellular immune function and hence virus activation among persons infected with herpes viruses suggests a suppression of cellular immune competence. Glaser and his colleagues have used increases in herpes antibody levels among seropositive persons as indicators of viral activation. In our study, we use HSV-1 antibody levels assessed prior to viral challenge. Serum samples were assayed for both HSV-1 IgG and HSV-1 IgA. HSV-1 antibody levels from HSV seropositive persons were (a) correlated with various psychological measures; (b) compared with nonspecific antibody levels, and with enumerations of peripheral white blood cell populations including lymphocyte

subsets; and (c) evaluated as a predictor of susceptibility to infection and clinical disease.

HSV Assays

To assay immunoglobulin against whole virus antigen an accredited HSV-1 strain was obtained (strain 10798 from the Centre for Applied Microbiological Research) and a pool of viral antigen prepared after growing the virus in human diploid fibroblasts. A control antigen pool was prepared from uninfected cell cultures. A pool of IgG-positive sera was also selected for use as a positive standard in the IgG assays. The format for the IgG ELISAs is similar to the protocol we employ for antirhinovirus antibody assays. One half of the microtitre plates are coated with viral antigen and the other half with control antigen. A titration of the positive serum standard and duplicate samples of each test serum (at a single dilution) are added to each half of the plate. After reading the final optical densities from the assays, the control antigen values are subtracted from the viral antigen values. The positive serum standard is assigned an arbitrary value of 1×10^5 units so the corrected optical densities from the test samples can be equated with a point on the standard dilution curve and allocated a value. We calibrate the positive serum standards against commercial sera of known immunoglobulin content, thus enabling the specific antibody content of the test-sera to be expressed in ng/ml.

The protocol adopted for HSV-1 IgA measurement utilized an indirect ELISA and test-sera pre-absorbed with protein-A. A suitable pool of IgA-positive sera was selected for use as a standard.

Of the 664 persons (with HSV assays), 82.7% were seropostive as assessed by HSV-IgA and 64.0% were serpositive as assessed by HSV-IgG. One hundred forty-two persons were seropositive on IgA but not on IgG. Conversely only 18 persons were seropositive on IgG but not on IgA. [Apparently there were differences in the sensitivity of our two assays]. As a consequence, seropositivity was defined as seropositive on either of the two measures; resulting in 569 [86%] seropositive persons.

Because increased antibody production to HSV-1 could reflect a general activation of humoral immunity as opposed to cellular immune suppression, we used anti-rubella antibody in test sera to control for this alternative interpretation. Necessary antigens were purchased from the Central Public Health Laboratory and their standard protocol adopted. Of the 569 persons seropositive on HSV-1, 96.0% were seropostive on Rubella.

HSV, Psychologic State, Immunity, and Susceptibility

Correlations with Psychologic Variables. Because we were concerned with associations between detectable HSV antibody levels and psychological variables, the following analyses included only those 569 persons who were seropositive on at least one of the two HSV

antibody measures.

Recall that serum samples for HSV antibody determination and psychological measures were both collected during the two day period prior to viral challenge. Cross-sectional correlations were calculated between HSV antibody levels (both IgA and IgG) and each of the following psychological measures: stressful life events, perceived stress, negative affect, personal control, self-esteem, role involvement in social networks (# of social roles), introversion-extroversion (I-E), and psychological distress. There were significant correlations between introversion-extroversion and IgA (.13, p<.002) and IgG (.12, p<.003). However, there was a similar correlation between Rubella antibodies and I-E (.14, p<.001) suggesting a general influence on antibody level rather than a specific influence on herpes antibodies. Similarly, there were significant correlations between number of social roles and IgA (.09, p<.03) and IgG (.08, p<.05), with a similar correlation between Rubella antibodies and social roles (.08, p<.06).

Correlations with Immune Measures. Correlations of HSV antibodies and immune measures (humoral immunity and cell enummerations) indicated HSV-IgA was correlated with serum IgA total (.09, p<.03), nasal IgA total (.12, p<.01), nasal protein total (.10, p<.02), and percent CD8+ cells (-.16, p<.04). HSV-IgG correlated only with percent CD8+ cells (-.18, p<.02). There were no correlations between Rubella antibody levels and any of these measures.

HSV antibodies as predictors of clinical colds. These analyses included the 249 subjects who were challenged with RV2, RV9, or RV14, were not administered a drug, and were seropositive on either HSV IgA or IgG. To control for virus, serostatus on the challenge virus, age, gender, education, allergy-status (allergic to something or not) and general (represented by Rubella) antibody level, each equation included each of these variables as well as the respective HSV antibody. The initial equations also included all possible interactions between the HSV antibody measure and control variables. Nonsignificant interactions were then dropped from the equation. There were interactions between virus type and IgG (coefficient of -.41, SE .19, t=-2.20, p<.028) and between virus type and IgA (coeff. of -.92, SE .40, t=-2.31, p<.02). Separate regressions within each virus indicate greater HSV-1 IgA antibody levels are associated with less risk for RV2 colds (coefficient -.66, SE .33, t=-2.01, p<.04), but not for RV9 or RV14 colds. In the case of volunteers exposed to RV2, 9.1% of those above the median developed colds, while 34.8% of those below the median developed colds. HSV-1 IgG was not associated with cold outcomes in any of the three viruses when examined separately.

Conclusion. We found no evidence for HSV antibody modulation by psychological variables. Although HSV-IgA antibody levels were related to susceptibility for RV2 colds, it was in the opposite direction than predicted. That is, persons with higher HSV-IgA levels were less susceptible to colds. We are, at this point, unable to explain why HSV antibody

levels would be negatively related to RV2 susceptibility and not related to susceptibility to RV9 or RV14. Both HSV-IgA and -IgG also demonstrated a small negative relation to the number of suppressor-cytotoxic cells (CD8+) in peripheral blood. Not surprisingly, HSV-IgA also demonstrated small positive correlations with measures of total Ig and total protein.

It is important that earlier studies demonstrated herpes antibody <u>change</u> between stressful and nonstressful situations. In our study, however, we were limited to cross-sectional correlations between a single measure of antibody and self-reported psychological variables. Individual variability in antibody production may overshadow any relation with psychosocial variables or with susceptibility to infection. Clearly, studies of herpes antibody changes in response to environmental stressors (e.g., medical school exams) provide a better test of this hypothesis.

The decreased risk for RV2 colds among persons with elevated HSV-IgA found in this study is puzzling. This may indicate that herpes antibody changes serve as a marker for more complex processes than a suppression of cellular immune function. Alternatively, it may suggest that the components of cellular function that are suppressed may have different implications in the face of different infectious agents.

Smoking, Immunity and Susceptibility to Colds

Most existing studies of smoking and infectious respiratory illness focus on influenza. For example, studies of the incidence of biologically verfied influenza during epidemics suggest greater risk of clinical illness among cigarette smokers (more than 1 pack/day: Finklea, Sandifer & Smith, 1969; at least 1/2 pack/day: Waldman et al., 1969). One study found that increased risk for smokers only occurred among those with little or no pre-epidemic antibody to the epidemic viruses (MacKenzie et al., 1976). However, the sole epidemiologic study of serologically verified rhinovirus colds (Gwaltney et al., 1966) failed to find a relation between smoking and disease risk. Finally, although data from the Tecumseh study (Monto et al., 1975) support greater incidence of a range of respiratory infections for smokers, effects of smoking on serologically verified illness rates varied with initial health status. Only the healthiest (<u>no</u> baseline symptoms of cough or sputum) group of smokers showed increased illness rates.

Although provocative, these studies suffer from several methodological weaknesses. First, smoking status and rate are based on self-report. Because smokers are often poor estimators of their smoking rates, vary in the way they smoke (e.g., deepth of inhaling, amount of the cigarette smoked) and sometimes bias reports made to health professionals, a biochemical assessment of rate and status is preferable. Second, exposure to the infectious agent is not controlled for in these studies. Smokers may be more (or less) likely to come in contact with infected persons. For example, smokers may have more children than

nonsmokers. Third, in most of these studies, the definition of an illness episode is dependent on persons seeking medical care (the exception is the Tecumseh study). Smokers may be more likely to seek medical care when bothered by mild symptoms because of their elevated risk for serious respiratory disease. Alternatively, they may be less likely to seek care because they fail to recognize mild respiratory symptoms as disease related.

In the study reported here, volunteer smoking status and rates are determined by serum cotinine--a metabolite of nicotine. Volunteers are exposed to one of three rhinoviruses and the onset and course of infection and symptoms are closely monitored during the study. Finally, diagnosis of clinical disease includes verification of infection as well as characteristic symptomatology.

We also examine the relationship between enumerations of peripheral white blood cell populations and smoking status and rate. We attempt to replicate earlier results indicating elevations across cell populations for smokers (see review by Holt, 1987) and examine the possibility that these changes predict susceptibility to rhinovirus colds.

Assessing Smoking Status and Rate

Serum cotinine. Cotinine, a metabolite of nicotine, was used to provide a biochemical assessment of smoking status and smoking rate. To obtain a representative and stable measure, we averaged cotinine levels from two serum samples taken four weeks apart. The first sample was collected at the Common Cold Unit at study intake and the second by volunteers' own physicians. Cotinine levels were measured in serum by gas chromatography (Feyerabend & Russell, 1990).

Smoking status. Persons with average cotinine levels of 15 ng/ml or more were defined as smokers, while those with less than 15 ng/ml were defined as nonsmokers (Jarvis, Tunstall-Pedoe, Feyerabend, Vesey & Saloojee, 1987). This cutoff is closely related to self-reports of smoking status when smokers are defined as persons smoking at least one cigarette per day. The use of a cotinine cutoff provides an objective means of classifying those whose self-reported rates are close to the criterion as well as those who may misrepresent their smoking status. Based on the 15 ng/ml coutoff, 9 persons (3% of the sample) who reported smoking less than one cigarette per day were defined as smokers, and 4 persons (1% of the sample) who reported smoking at least one cigarette per day were defined as nonsmokers.

Smoking rate. We use log average cotinine levels as an indicator of smoking rate. Cotinine is used because it provides an objective measure of nicotine intake that is not subject to self-report bias. Cotinine also provides a comparable measure for cigar and pipe smokers. The correlation between log average cotinine and the log self-reported number of cigarettes smoked per day (including nonsmokers) was .96 (p < .001, N = 302). The same correlation for smokers only is .54 (p < .001, N = 83).

Smoking and Enumerations of Peripheral White Blood Cell Populations

Table 1 presents the mean enumerations of peripheral leukocyte population by smoking status: never smokers, 5 years or less ago, current light (230 ng/ml or less) and heavy (more than 230 ng/ml) smokers. The 230 ng/ml cutoff is based on the median average cotinine level for 211 smokers included in total white blood cell analysis and is equivalent to smoking about 15 cigarettes per day. The probabilities in the table are derived from analyses of covariance in which the independent variable was smoking status (5 levels); the dependent variables were the $\log_{10}$ transformed enumerations of cell populations; and the covariates (control variables) were sex, age, education, and allergy status. For interpretability, the table reports observed mean cell enumberations rather than their logs.

Table 1. Mean enumerations of peripheral white blood cell populations by smoking status. All values are 10^6/ml.

	Never-Smokers	Ex-Smoker >5 years	Ex-Smoker <5 years	Current light	Current heavy	p<
Total WBC	5.56	5.74	6.32	6.43	7.65	.001
Monocytes	.45	.48	.51	.53	.60	.001
Neutrophils	3.20	3.32	3.80	3.71	4.59	.001
Lymphocytes	1.90	1.94	2.01	2.19	2.45	.001
T-cells	1.39	1.36	1.72	1.60	1.96	.004
B-cells	.30	.29	.34	.35	.38	NS
Helpers	.68	.68	.84	.76	1.05	.037
Supp.-Cytot.	.31	.27	.34	.33	.46	.093

Sample sizes for monocytes, lymphocytes and neutrophils are approximately 348 for never-smokers, 74 for ex-smokers who quit more than 5 years ago, 54 for ex-smokers who quit 5 years or less ago, 102 for current smokers with average cotinine levels of 230 ng/ml or less, and 109 for current smokers with average cotinine levels of more than 230 ng/ml. Sample sizes for T, B, helper and suppressor-cytotoxic cells are approximately 88, 22, 17, 22, and 21.

There is a clear pattern across cell populations with smokers having more cells than nonsmokers and heavy smokers having the greatest number of cells. Although the patterns look similar, the differences do not reach significance among B-cells and suppressor-cytotoxic Ts. In both of these cases the sample sizes are considerably smaller than those available for WBC, monocytes, neutrophils, and lymphocytes and hence statistical power is much attenuated. These data are consistent with those reported earlier by a number of investigators (see review by Holt, 1987).

Tollerud et al. (1989) report that elevated numbers of white blood cells among smokers

are reversible with persons quitting one-year or more prior to the study having counts like nonsmokers. Our own data indicate that persons quit for less than five years have mean cell numbers similar to those of light smokers. Only after five years off cigarettes do their means resemble those of never smokers.

Smoking and Susceptibility to Rhinovirus Colds

Regression Equations Testing the Effects of Smoking on Susceptibility to Colds

The primary outcome in these analyses is whether or not persons exposed to viruses developed clinical colds (verified infection + clinician diagnosis of a cold). In each case, we also conduct additional analyses to determine whether effects on clinical colds are attributable to increased infection or to an increase in clinical colds among infected persons. Logistic regression is used to predict all of these binary outcomes. Multiple regression is used in supplementary analyses in which we predict total mucus weights and total symptom scores.

Predictors in the prototypic regression included smoking (status or rate) as well as control variables that might provide alternative explanations for the effect of smoking. The controls included age, gender, education, allergies, virus, and serostatus. The categorical variables, virus (RV2, RV9, RV14), serostatus, allergies (yes or no) and gender were entered as dummy variables, while age and education (no schooling [0] to higher degree [8]) were entered as continuous variables. Preliminary analyses were conducted to determine whether there were interactions between any of the control variables and smoking in predicting the outcome under consideration (see Hosmer & Lemeshow, 1989). As discussed later, a smoking by gender interaction was detected in all of the analyses; there were no significant interactions between smoking and any other control variable. There were also no three-way interactions of control variables, gender and smoking. The lack of interactions with other control variables indicate that reported effects are similar across different viruses, and levels of serostatus, age, allergy status, and education. Hence the final (reported) equations included all of the control variables, smoking (either status or rate), and the smoking by gender interaction. As a result, regression coefficients for smoking and smoking by gender are adjusted for possible effects of the control variables.

All reported rates of infections and colds are adjusted (predicted rates) for all variables in an equation. In the case of interactions between smoking and gender, predicted rates, adjusted coefficients, and odds ratios were derived from separate equations in which the effects of smoking were examined separately for males and females.

Smoking as a Predictor of Clinical Colds

Smoking status was entered as dichotomized variables in the initial equation predicting clinical colds. There was a significant coefficient for the smoking by gender interaction (coefficient of 1.25, standard error of .61, $t = 2.05$, $p < .04$). To accurately estimate the nature of the interaction, we calculated separate regression equations for men and for women. Coefficients and odds ratios for smoking status and adjusted (predicted) clinical cold rates were derived from these regressions. For women, 31.7% of nonsmokers and 56.2% of smokers developed clinical colds. The coefficient for smoking status was 1.47, standard error .51, $t = 2.89$, $p < .004$; the odds ratio was 4.33. The odds ratio of 4.33 indicates that women smokers were 4.33 times more likely to develop colds than women who did not smoke. For men, 31.2% of smokers and 29.4% of nonsmokers developed colds. This difference was not statistically reliable.

One explanation for smoking status influencing risk of colds for women but not for men would be that smoking rate differed between men and women smokers. For example, one might hypothesize that women smokers smoked more and that higher smoking rates were associated with higher risk. However, t test comparing mean self-reported rates (16.1 for men and 15.2 for women) and mean log averaged cotinines (2.27 for men and 2.27 for women) indicate no differences between men and women. Hence increased risk of developing colds for women smokers is not attributable to their being heavier (or lighter) smokers.

A second series of analyses was designed to determine the possibility of a dose-response relation between smoking rate and incidence of colds. In these analyses, smoking rate (continuous log average cotinine levels) was entered into the standard logistic model in place of smoking status. The analysis indicated a significant coefficient for the smoking rate by gender interaction (.69, standard error of .30, $t = 2.27$, $p < .02$). The nature of this relation is depicted in Figure 1. Three cotinine levels are used to depict rate in the figure: nonsmokers (15 ng/ml or less), and light (15.01 to 203.17 ng/ml) and heavy (203.18 or higher ng/ml) smokers. The light and heavy smoker categories are based on a median split of average cotinine for the 83 smokers in the sample of 302 persons used in these analyses and correspond to 14 or fewer cigarettes per day and more than 14 cigarettes per day respectively. Standard errors of each of these rates are also depicted in the figure. As apparent from the figure, women smokers are at higher risk for developing colds than women nonsmokers and that risk increases with increased smoking rate. However, smoking does not increase risk for men.
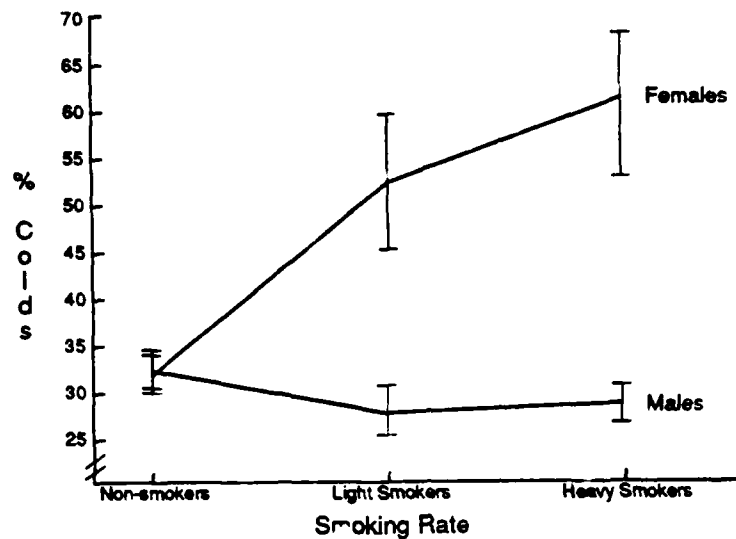
Figure 1. Smoking rate (based on average cotinine) and gender as predictors of the percent of persons developing a clinical cold.

Infection. Additional analyses were conducted to determine whether the previously described effects were attributable to the influence of smoking on increased infections or on increased clinical colds among infected persons. In predicting infection, smoking status, gender, and the smoking status by gender interaction all failed to reach levels of significance. A similar analysis using smoking rate (log average cotinine) also failed to indicate any significant effects.

Predicting clinical colds for infected persons. A regression predicting clinical colds for infected persons (N = 246) produced a significant coefficient for the smoking status by gender interaction (1.69, .68, t = 2.49, p < .01). Separate regressions models for men and women indicated a significant coefficient for smoking status for women (1.81, .59, t = 3.04, p < .002) but not for men. The odds ratio was 6.10 indicating that infected women who smoked were more than six times as likely to develop clinical colds than infected women who did not smoke. A similar analysis using smoking rate (average cotinine) also indicated a rate by gender interaction (.88, .33, t = 2.66, p < .008). The effect, depicted in Figure 2, mapped perfectly on to the modifying influence of smoking status on clinical colds for women.
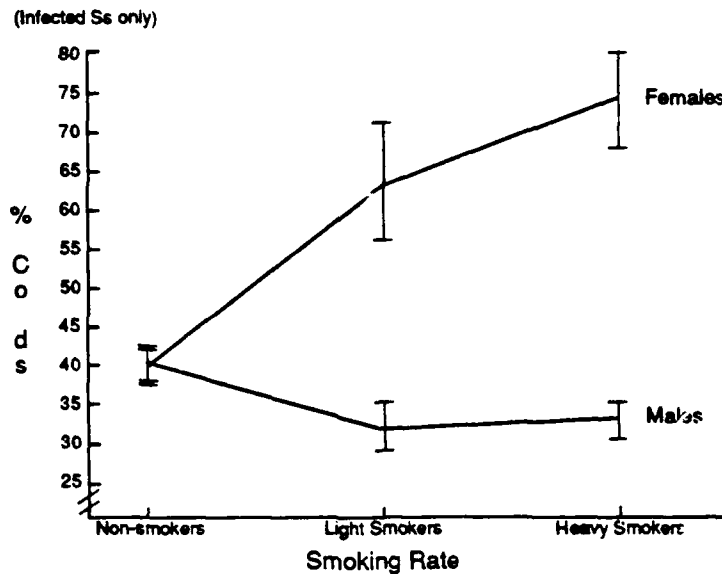
(Infected Ss only)

Figure 2. Smoking rate (based on average cotinine) and gender as predictors of the percent of clinical colds among infected persons (those shedding virus or antibody increase).

The smoking rate by gender interaction depicted in Figure 2 seems to provide the best explanation of the influence of smoking on clinical colds for women. The data suggest that the interaction is attributable to clinical symptomatology among infected persons rather than differences in infection rates.

Smoking Status as a Predictor of Symptoms and Mucus Weights

As discussed earlier, the definition of clinical colds includes biological verification of infection (virus shedding and/or antibody response) and the clinician's judgment that the volunteer has a clinical cold. We also examined two continuous indicators of cold symptomatology: the total symptom score and total mucus weights. Because symptom scores and mucus weights are measures of signs and symptoms but not infection, only infected persons (N = 246) were included in these analyses.

Symptom scores derive from a standard respiratory symptom protocol (described by Beare and Reed, 1977). Mucus weights are determined by collecting tissues used by volunteers in sealed plastic bags. The bags are weighed and the weight of the tissues and bags are subtracted. Mucus weights are not used in calculating symptom scores. Total symptom and total mucus scores are calculated for both pre- and post-challenge. The pre-challenge measures are based on the sum of the two days prior to challenge. The mode pre-challenge score in both cases was 0. The post-challenge measures are based on the sums of daily scores and weights from days 2 through 6 post-challenge. All of these scores

were $\log_{10}$ transformed to normalize the distributions.

A linear multiple regression model was used in analyzing these continuous data. Procedures and models are the same as those described earlier. First, we examined the possibility of a relation between smoking and pre-challenge (baseline) symptom scores and mucus weights. There were no relations of smoking, gender or their interaction and either pre-challenge score. Second, we examined the relations of smoking, gender and their interaction in predicting five-day post-challenge symptoms and mucus weights. In the case of total symptoms, there was a smoking status X gender interaction (F[1,236] = 9.28, p < .003). The smoking rate analysis indicated a similar interaction (F[1,236] = 10.23, p < .002). As apparent from Figure 3, these data are consistent with the clinical cold data based on infection and clinician judgment.
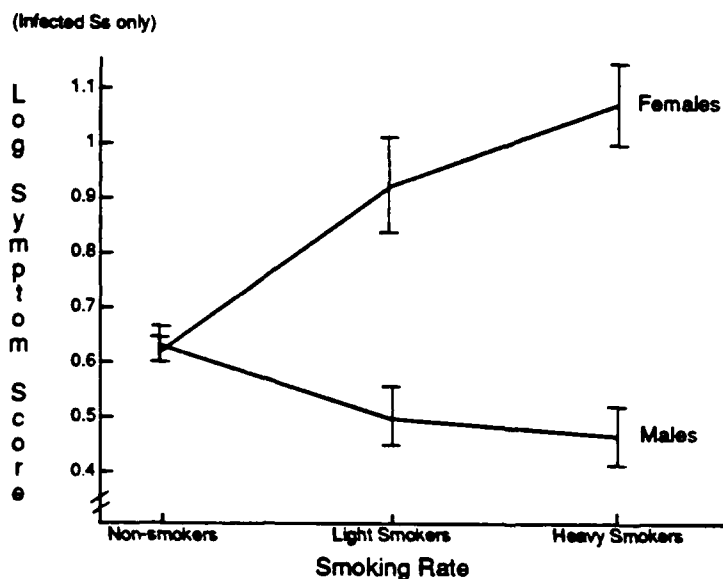

Figure 3. Smoking rate (based on average cotinine) and gender as predictors of log total symptom scores among infected persons.

As apparent from Figure 4, virtually identical results were found for mucus weights. In this case, there was a main effect for smoking status (F[1,237] = 4.49, p < .035) and a smoking status by gender interaction similar to that found in the earlier analyses (F[1,236] = 9.63, p < .002). For smoking rate, there was similarly an effect for rate (F[1,237] = 4.02, p < .046) and a rate by gender interaction (F[1,236] = 10.59, p < .001).
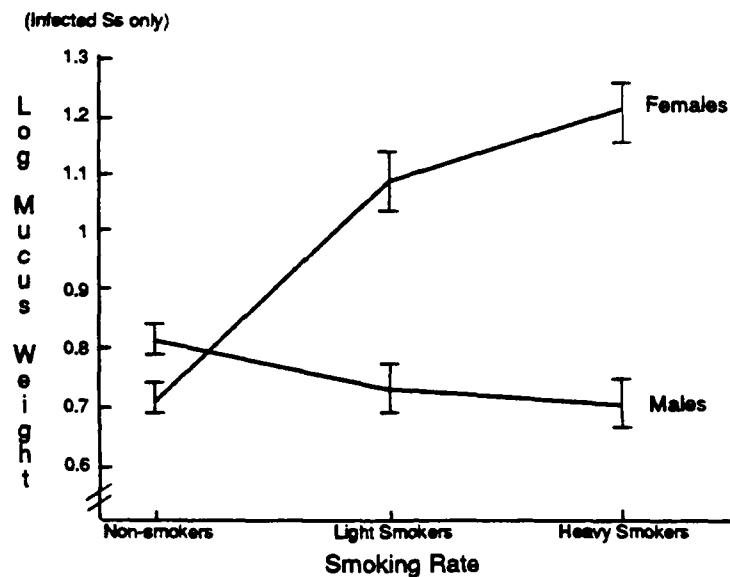
Figure 4. Smoking rate (based on average cotinine) and gender as predictors of total mucus weights among infected persons.

Pathways through which Smoking may Influence Susceptibility

Our data indicate that smoking puts women but not men at higher risk for the development of clinical colds when exposed to a rhinovirus. Moreover, the influence of smoking on increased susceptibility in women primarily occurs at the level of symptom development for infected persons. We also find increased numbers of peripheral blood leukocytes among smokers. Although we found no smoking by gender interactions in predicting leukocyte numbers, mean increases under smoking tend to be slightly higher for females than males (cf. Burton et al., 1983). Even so, these two effects seem to be independent of one another since adding leucocyte numbers to the equation predicting susceptibility to colds has only a trivial influence on the outcome.

Smoking and susceptibility to illness. The increased risk of clinical illness for women smokers is generally consistent with the epidemiologic data reviewed earlier in this report. What is puzzling is why women but not men are affected. Because we assess smoking status and rate biochemically, gender differences in the manner in which cigarettes are smoked do not provide an explanation. Our data are, however, consistent with accumulating evidence that females are more sensitive to nicotine than males (Grunberg, in press; Silverstein et al., 1980). For example, there is evidence from human studies that the first experience with cigarette smoke is more aversive for women than men (e.g., Silverstein, 1980) and from animal studies that there are greater effects of nicotine on eating behaviors and body weights of female rats (Grunberg, Bowen & Winders, 1986). More closely tied to

the issue of smoker risk for infectious respiratory illness is evidence for sex-related differences in effects of smoking on lung function. Although the data are not entirely consistent (USDHHS, 1984), women smokers (but not nonsmokers) may have higher prevalence of abnormalities in small airways response than men (e.g., Buist & Ross, 1973a; 1973b; Buist et al., 1973; 1979a). These responses are thought to be irritative.

Smoking and symptom mediators. We can only speculate on why women smokers are at higher risk for colds and men are not. However, whatever the gender difference contributing to this effect, it must influence symptom production for infected persons. Symptom manifestation may be influenced by a variety of mechanisms. The production of more virus may itself account for increased symptomatology ⋅ may trigger the release of symptom mediators. A rough measure of viral replication, number of days shedding virus, did not however map onto the gender by smoking effect. Alternatively, differences between male and female smokers in the production and/or binding of symptom mediators such as kinins or histamines may be responsible. Rhinovirus infections are presumed to trigger a process that results in the production of inflammatory mediators (Naclerio et al, 1988). These mediators interact with the local environment in the nose and throat to cause rhinorrhea, nasal congestion and other cold symptoms (Gwaltney et al., 1984). Recent evidence indicates that kinins are a probable mediator with increased kinins found in the nasal secretions of persons developing experimentally induced colds (Naclerio et al., 1988) and nasal obstruction, sore throats and rhinorrhea found among human volunteers challenged with bradykinin (Proud et al., 1987). Enzymes with arginine esterase activity (probably plasma kallikrein) are responsible for generating kinins (Naclerio et al., 1988). Although Gwaltney and his colleagues (e.g., Naclerio et al., 1900) have been unable to detect histamines release in persons with rhinovirus colds, evidence for symptom suppression in response to nedocromil (Barrow et al., 1990), a drug that inhibits mast cell degranulation, suggests that histamines and leukotrienes may also be involved in symptom mediation. Finally, the direct irritation of effected membranes (see discussion of small airways above) may lower the threshold for or aggravate existing symtomatology.

Acute versus chronic effects. Volunteers were allowed to smoke during the trials. As a result, it is unclear whether the effects we report are acute effects on host resistance during viral challenge or chronic effects of smoking.

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